a homogenate of whole thymus, do not belong to the lymphoid cells of the gland. These workers also showed that in vitro thymocytes do not exhibit sensitivity for androgens.

We concluded that the thymolytic action of androgenic-anabolic steroids is not the result of their direct effect on thymocytes, but is realized indirectly by endogenous factors controlling metabolic processes in lymphoid tissue.

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EFFECT OF UHF RADIATION AND EXPOSURE TO COLD ON DESTRUCTION AND REPAIR OF REGENERATING BONE IN DOGS

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Local cryosurgical destruction combined with physical and chemical factors, namely laser therapy and ultrasound, is used in organ-conserving operations of a sufficiently radical nature in oncology [2, 3, 5]. The mechanism of action of ultrahigh frequencies (UHF) is not always the same, but most investigators [1, 4] assess its effect as potentiating the destructive action of cold. Under the influence of UHF microwaves the thermophysical characteristics of biological tissue are modified, and this leads to a greater increase in the volume of cryonecrosis. Morphological changes in bone tissue after exposure to cold and in combination with UHF radiation have not been studied. We have investigated the character of prophylactic changes in tissues during cryosurgery, alone and in conjunction with preliminary UHF irradiation, with attention to the character of repair processes. The experimental model was mechanical injury to the mandible inflicted by a drill, allowing the proliferative reaction of cells of osteogenic tissue, which simulates to some degree the changes of neoplastic cell proliferation, to be studied.

EXPERIMENTAL METHOD

Altogether 36 experiments (four series) were carried out on the mandible of 18 mongrel dogs, bilaterally. The operations were performed under hexobarbital anesthesia. Series I (control group) involved mechanical injury to the body of the mandible without any additional procedures; in series I the bone wound was irradiated by UHF radiation 3 days after its formation; in series III the bone wound was treated cryosurgically 3 days after its formation; in series IV cryosurgery was preceded by UHF irradiation; 3 days after formation of the bone wound also.

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After aseptic treatment of the operative field a skin incision 5 cm long was made in the submandibular region on both sides of the bone, after preliminary stripping of the periosteum. A burr-hold was drilled in the outer cortical lamina 0.5-0.6 cm from the upper edge of the mandible, forming a bone wound 0.5 cm deep and 0.3 cm in diameter perpendicularly to the body of the mandible. An operative channel was thus formed. The cortical lamina on the lingual aspect remained intact. The wound was sutured. Three days later the experimental animals were treated by cryosurgery, either alone or preceded by UHF irradiation; under anesthesia and after retraction of the wound edges along the previous incisions, the vestibular surface of the body of the mandible was exposed in the region of the previous operative trauma. The procedure was planned so that the entrance to the burr-hole was located in the center of the cryosurgical applicator or the UHF source (model Luch-2). The optimal duration of irradiation up to a temperature of 41-42°C was 6-8 min, its power 10 W, and frequency 2375 MHz.

An independent applicator with a flat circular tip 1.5 cm in diameter was used for cryosurgery. The temperature of the tip was −100°C and the duration of its action 12 min. Under these conditions the linear diameter of the zone of freezing averaged 3.0 cm. The soft tissues surrounding the mandible were carefully isolated from damage and were sutured in layers after the end of the operation. The animals were killed by an overdose of hexobarbital on the 6th and 8th days after wound formation and on the 3rd and 15th days after the different forms of treatment in the experimental groups. Thus four animals were used at each stage of observation.

Material for histological investigation was prepared as follows. A total preparation of the mandible, including not only the body but also the alveolar process with teeth, with a total length of 3.5 cm along the horizontal axis, was excised in the zone of the bone wound and subsequent treatment. The tissue block comprised the bone wound, the whole zone of freezing, and the adjacent zone of surrounding bone tissue within a radius of 3-4 mm. After decalcification, this fragment was divided into three segments, which were dehydrated with alcohols and embedded in celloidin. Sections 30 μm thick were stained with hematoxylin and eosin (750) and by Van Gieson's method.

EXPERIMENTAL RESULTS

In the control series of experiments 6 days after the operation (Fig. 1a) signs of serious inflammation and hemorrhagic foci were observed in the lumen of the operative channel and adjacent medullary spaces in its wall, accompanied by areas of fibroreticular tissue, detected mainly in the adjacent medullary spaces. No staining of osteocyte nuclei was present in the bone tissue in the walls of the operative channel. Yellow marrow with tiny hemorrhages and tissue cysts was present in the intertrabecular spaces of the bone tissue of the mandibular canal.

In series II, 3 days after UHF irradiation (6 days after formation of the bone wound; Fig. 1b) nuclei of osteocytes of bone tissue forming the wall of the operative channel and in the lumen of the channel did not stain. If serious inflammation was present and there were remnants of blood clot in the lumen of the channel a zone of fibroreticular tissue could be seen and was much greater than in the control. The intertrabecular spaces were filled with undifferentiated fibrocellular tissue, rich in blood vessels.

In series III (Fig. 1c), 3 days after exposure to cold, tissue detritus was observed in the operative channel and also in the mandibular canal. Edematous yellow marrow with numerous tissue cysts and small hemorrhagic foci were present in the medullary spaces around the operative channel and in more distant zones. Bone trabeculae at a distance from the channel were indented and reduced in thickness, so that the medullary spaces appeared widened.

In series IV (Fig. 1d), 3 days after a combination of UHF irradiation and cryosurgery (6 days after formation of the bone wound) signs of serious inflammation were present in the operative channel, just as in the control series of experiments. Medullary spaces in the bone tissue of its walls were widened and contained fibrocellular tissue, and some of the blood vessels were of the sinusoidal type. The outlines of individual bone structures were uneven because of absorption of bone substance. The mandibular canal and medullary spaces at a distance from the bone wound were partly filled with edematous yellow marrow and fibrocellular tissue.

After 18 days, in series I (the control group) the lumen of the operative channel contained not only fibroreticular tissue and single developing bone trabeculae, but also a small residual focus of serious inflammation. Indentations were visible in the trabeculae of the cancellous substance in the walls of the channel. In the zone of detachment of the periosteum in connection with the drilling, moderately severe reabsorption was visible on the periosteal surface of the body of the mandible. The outer layers of the periosteum were restored above the entrance to the channel, and proliferation was present.

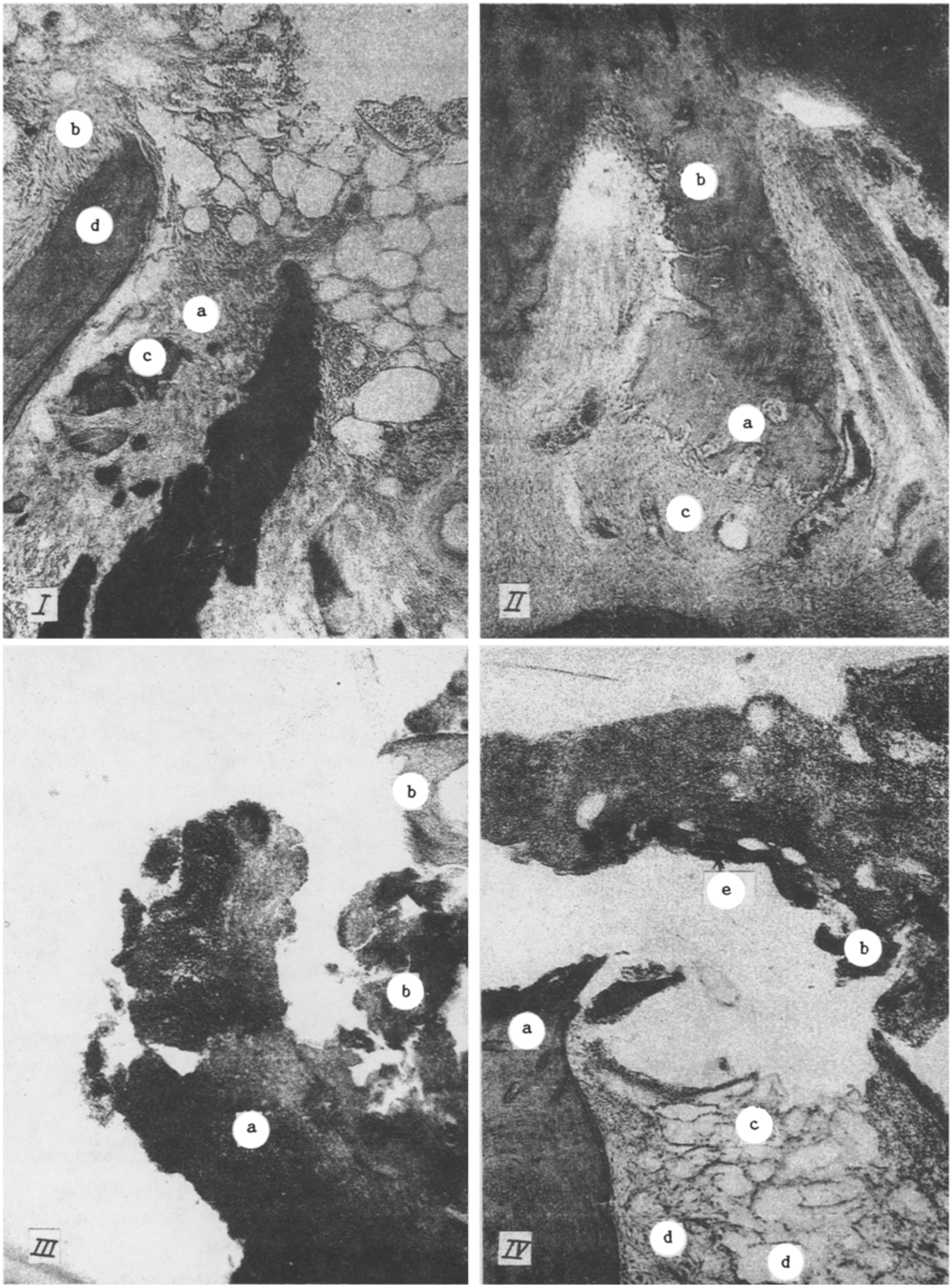

Fig. 1. Morphological changes in regenerating bone in control (I), after UHF irradiation (II), after cryosurgery (III), and after a combination of UHF and cryosurgery (IV). I) Six-day regenerating bone; a) leukocytes, b) fibrocellular tissue, c) small spicules of bone; d) bone trabecula; II) 3 days after UHF irradiation of 3-day regenerating bone: a) newly formed bone trabeculae, b) pre-existing bone structures, c) fibroreticular osteoblastic tissue; III) 3 days after cryosurgery on 3-day regenerating bone: a) tissue detritus, b) remnants of bone trabeculae; IV) 3 days after combined UHF and cryosurgery on 3-day regenerating bone: a) bony wall of channel, b) remnants of bone trabeculae, c) disintegrating bone marrow, d) fibrocellular tissue, e) tissue detritus.

In series II, 15 days after UHF irradiation (18 days after formation of the bone wound) fibroreticular tissue with immature bone trabeculae could be seen in the lumen of the operative channel. In the cancellous substance of the channel walls there were many instances of the formation of new layers of bony trabeculae on old, pre-existing layers. The intertrabecular spaces remote from the bone wound, and also the mandibular canal were filled with fibroreticular tissue. Solitary immature bone trabeculae were present in the walls of the canal. On the periosteal surface of the body of the mandible, at a distance from the bone wound, marked evidence of bone formation was present in the form of a dense network of immature bone trabeculae.

In series III, 15 days after cryosurgery (18 days after formation of the bone wound) localized inflammatory infiltration could be seen in the lumen of the operative channel, together with polymorphonuclear leukocytes. Edematous fibrocellular tissue with tissue cysts and single newly formed bone trabeculae were present in the intrabecular spaces of the cancellous substance at a distance from the wound and in the mandibular canal. Considerable restructuring of bone tissue was observed in zones remote from the site of operation. Moderately intensive reactive bone formation was present on the periosteal surface at a distance from the operative channel, and indentations were seen close to its entrance.

In series IV, 15 days after cryosurgery preceded by UHF irradiation (18 days after creation of the bone wound) moderately intensive local productive inflammation with tissue detritus could be seen in the lumen of the channel. Yellow bone marrow with moderately severe edema, and a mild degree of bone tissue resorption in the form of indentations, were visible in the diluted medullary spaces of the cancellous substance in the walls of the operative channel and the mandibular canal. The periosteal surface of the body of the mandible a short distance from the bone wound showed small indentations, whereas at a distance, periosteal bone formation could be seen.

Thus exposure to cold induces marked necrobiotic changes in the proliferating tissue of the regenerating bone, formed in the bone wound and tissues of the medullary spaces around the wound, with stimulation of repair processes in the bone tissue away from the site of cryosurgical application. This effect of cold on proliferating tissue indicates that it may be used during removal of tumors to allow extensive excisions and resections to be replaced by more or less conservative operations: by a combination of excision of the tumor and cryosurgical destruction of the surrounding tissue. The combination of cold and UHF radiation alleviated to some degree the adverse action of cold on the bone tissue away from the wound, in the form of resorption, and reparative processes in the tissues remaining after destruction could be seen to be proceeding more intensively and on that basis this method can be recommended for clinical use in the treatment of benign tumors.

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